

WHEY PROTEIN HYDROLYSATES AND MIXTURES THEREOF WITH CASEIN AND/OR SOY PROTEIN HYDROLYSATES

This is a continuation of application Ser. No. 08/353,652 filed Dec. 9, 1994, now abandoned, which in turn is a continuation of application Ser. No. 07/960,143, filed Oct. 13, 1992, now abandoned, which in turn is a continuation of application Ser. No. 07/591,593, filed Oct. 2, 1990, now abandoned.

The invention provides a controlled protein hydrolysate from cow milk proteins and soy proteins, a process for preparing such hydrolysate and compositions comprising such hydrolysate.

Protein hydrolysates may be used in food for dietary or therapeutic purposes. It is generally accepted that the size of peptides should be small and that the content of free amino acids should be low, to secure optimum absorption by the enteral tract.

Various protein hydrolysates and processes for their manufacture have been suggested. They all show certain drawbacks, with respect to their composition, their manufacture, or both.

The present invention provides an outbalanced and controlled mixture of physiological small peptides which secures optimal absorption and which is moreover free of allergenic and preserves the structures susceptible of exerting anticipatory regulations to maximize the tolerance and protein metabolism and the structure of other biologically active natural peptides.

Such mixture can be obtained employing selected starting materials, enzymes and reaction steps.

The present invention provides a protein hydrolysate of whey protein substantially free of proteins having a molecular weight of more than 60,000.

The whey protein fraction substantially free of proteins having a molecular weight of more than 60,000 is herein-after designated selected whey protein.

A preferred selected whey protein hydrolysate of the invention is obtained by physiological hydrolysis, involving a gastric phase, i.e. a HCl/pepsin prehydrolysis, followed by an enzymatic treatment of the prehydrolysate with a mixture of trypsin-chymotrypsin with a cationic serine endoprotease type elastase 2.

The introduction of a gastric phase and the use of cationic serine endoprotease type elastase 2, preferably such hydrolyzing at P1 methionine and P1 leucine residues in the whey protein hydrolysis procedure, allow the denaturation of globular proteins such as albumine or globulines and of tertiary and quaternary structures. Such denaturation does not only allow the elimination of allergenic properties but also facilitates the access by the pancreatic enzymes trypsin-chymotrypsin to hydrolysis sites, while preserving the physiological peptide sequences susceptible to induce the reactions of anticipatory regulations and other regulatory functions.

The term cationic serine endoprotease type elastase 2 as used herein relates to enzymes capable of hydrolyzing under alkaline conditions the peptide bonds formed between leucine, phenylalanine, methionine and tyrosine with glycine and alanine and include for example porcine elastase type 2, human pancreatic elastase type 2, and cationic serine endopeptidases having a similar activity.

The whey protein used as starting material for the preparation of the whey protein hydrolysate may be obtained by removal of the proteins and macropeptides present in the protein whey fraction in a manner known per se in the art,

e.g. employing anion-exchange resins, but preferably by micro- or ultrafiltration, employing conventional membranes having the required dynamic cut-off capacity.

The whey protein fraction employed will conveniently be substantially free of macrolipids to facilitate enzymatic hydrolysis. (The term macrolipids as used herein refers to the residual milk fat material finely emulsified in the form of microglobules of triglycerides and polar lipids of the phospholipid and lipoprotein type.) Such macrolipids may be removed prior to the removal of the proteins and macropeptides or simultaneously, e.g. by micro- and/or ultrafiltration. In general it will be preferred to prepare the selected whey protein fraction employed starting from whey protein comprising macrolipids and to subject this material to an ultra- or diafiltration. This procedure has the advantage that it employs membranes having a cut-off capacity of more than 500,000, e.g. of 1,000,000 while resulting in an effective or dynamic cut-off of 50,000 to 100,000.

Depending on the desired qualities of the whey protein fraction employed, the ultrafiltration will be carried out with membranes having a dynamic cut-off capacity in the range of from about 50,000 to about 1,000,000, preferably of about 50,000. The term dynamic cut-off capacity as used herein is defined as the cut-off observed after a time of stabilization of the dynamic membrane when ultrafiltrating whey protein solution.

The selected whey protein used as starting material may be delactosed (or not), depending on the contemplated use of the hydrolysate. It is preferably delactosed.

A preferred hydrolysis process of the selected whey protein involves typically the steps of

a) heating a solution of selected whey protein in water to $43^{\circ}\pm 4^{\circ}$ C., and subjecting said solution to a pepsin prehydrolysis at pH between 2.0 and 3.0,

b) adjusting the pH of the mixture of step a) at a temperature in the range of from 35° to 50° C. to a pH between 7.0 and 9.0 and submitting said mixture to an enzymatic trypsin-chymotrypsin hydrolysis in the presence of a cationic serine endoprotease of type elastase 2,

c) pasteurizing the mixture of step b), subjecting it to an ultrafiltration and concentrating and drying the permeate.

The pepsin prehydrolysis according to step a) is conveniently effected with an extract of bovine pepsin, obtainable by extraction of the rennet-stomach of adult cows. The pepsin extract preferably comprises at least 1700 mg pepsin per litre. The ratio of the mass of active chymosin: mass of active bovine pepsin is preferably equal to or smaller than 0.154. The weight ratio pepsin: substrate is conveniently in the range of 1:7500 to 1:2500, preferably about 1:5000. The prehydrolysis is preferably carried out with agitation, more preferably with rhythmic controlled agitation simulating the peristalsis of the stomach.

Where desired, the prehydrolysate is demineralized before subjecting it to the enzymatic hydrolysis of step b). The demineralization may be effected in a manner known per se for the reduction of the chloride, sodium and potassium content, e.g. by ultrafiltration employing membranes allowing to retain a maximum of peptides. Suitable membranes have a cut-off of from 1500 to 15000 e.g. of ca. 10,000. The pH of the prehydrolysate is conveniently adjusted to pH 8 ± 0.1 in a manner known per se. A suitable ultrafiltration temperature is between 30° to 60° e.g. ca. 50° C.; a suitable inlet pressure is conveniently between 2 and 4 bars.

The optionally demineralized mixture of step a) may be rendered alkaline in a manner known per se, e.g. employing